# Equations Predict PER From Amino Acid Analysis

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□ NUTRITION-CONSCIOUS CONSUMERS and industry are becoming increasingly aware that there is more to protein than just quantity, and that certain foods are of higher quality than others. Also, government is starting to require that food ingredients replacing other ingredients in formulations may be used only if the finished food protein quality is not lowered significantly. Thus, protein quality must be measured.

Established methods for the measurement of protein quality are few in number and are usually quite expensive, because most are time-consuming biological assays. The most widely used method is the protein efficiency ratio (PER) test, which measures protein quality by feeding a diet containing 10% of the test protein to weanling rats for 28 days and measuring their weight gain; the PER is the weight gained by the rats divided by the weight of protein consumed (AOAC, 1970). The cost for a single sample assay varies between \$200 and \$500, and the total elapsed time is about 45 days. The expense and time required for the assay make it impractical for quality control

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and regulatory control of food products.

Several food standard proposals by the U.S. Department of Agriculture require a demonstration of adequate protein quality, but PER is the only method that has been accepted by the Association of Official Analytical Chemists (AOAC). Therefore, it would be desirable to develop a technique for estimating PER that is less expensive and more rapid. It seems reasonable to assume that the relative quantities of the various amino acids in the food could be used as reliable estimators of actual protein quality. Amino acid analysis is reasonably inexpensive (\$60-\$100), and results can be available between 24 and 48 hr.

A study was therefore undertaken to determine whether regression equations could be developed that would effectively predict the expected protein efficiency ratio from an amino acid analysis.

### SAMPLES PREPARED & ANALYZED

Eight samples of beef tissue—one sample of eye of round, three samples of partially defatted beef fatty tissue, two samples of partially defatted chopped beef, one sample of cooked cured partially defatted chopped beef, and one sample of collagen derived from beef hide—were selected to be fed to rats following the AOAC prescribed method for PER as first described by Derse

Table 1—CORRELATION COEFFICIENTS between amino acids

	HIS	ILE	LEU	LYS	MET	CYSH	PHE	TYR	THR	TRY
HIS		0.944	0.973	0.957	0.907	0.695	0.949	0.971	0.952	0.881
ILE	0.944		0.960	0.981	0.983	0.603	0.911	0.985	0.691	0.718
LEU	0.973	0.960		0.982	0.837	0.491	0.999	0.982	0.764	0.794
LYS	0.957	0.981	0.982		0.926	0.423	0.946	0.999	0.852	0.731
MET	0.907	0.983	0.837	0.926		0.269	0.800	0.852	0.852	0.654
CYSH	0.695	0.603	0.491	0.423	0.269		0.454	0.423	0.346	0.360
PHE	0.949	0.911	0.999	0.946	0.800	0.454		0.982	0.764	0.794
TYR	0.971	0.985	0.982	0.999	0.852	0.423	0.982		0.852	0.731
THR	0.952	0.691	0.764	0.852	0.852	0.346	0.764	0.852	- <del></del> -	0.808
TRY	0.881	0.718	0.794	0.731	0.654	0.360	0.794	0.731	0.808	<del></del> . 75
VAL	0.650	0.265	0.340	0.269	0.115	0.999	0.340	0.269	0.192	0.200
ALA	-0.436	-0.714	-0.357	-0.800	-0.873	-0.076	-0.643	-0.727	-0.800	-0.886
ARG	-0.218	-0.214	-0.571	-0.291	-0.218	-0.756	-0.286	-0.218	-0.291	-0.322
ASP	0.582	0.714	0.500	0.946	0.873	0.227	0.786	0.873	0.946	1.000
GLU	0.727	0.714	0.500	0.946	0.873	0.378	0.786	0.873	0.946	1.000
GLY	-0.727	-0.857	-0.643	-0.946	-0.873	-0.378	-0.929	1.000	-0.800	-0.886
HYLYS	-0.481	-0.618	-0.837	-0.778	-0.704	-0.577	-0.691	-0.778	-0.630	-0.698
HYPRO	-0.873	-0.857	-0.643	-0.800	-0.727	-0.529	-0.929	-0.873	-0.800	-0.886
PRO	-0.800	-0.786	-0.571	-0.873	-0.800	-0.454	-0.857	-0.800	-0.873	-0.467
SER	0.528	0.593	0.222	0.679	0.755	0.078	0.519	0.604	0.830	0.920

(1965). Samples of partially defatted beef products were received frozen from several establishments preparing these products. After initial proximate analysis of the beef tissue, samples were freeze-dried, and proximate analysis and amino acid assay were conducted (Happich et al., 1974). Fat, moisture, and total nitrogen determinations were accomplished using standard methods for meat and meat products (AOAC, 1970).

Amino acid analyses were determined by use of a Piez-Morris ion-exchange column after sample hydrolysis with 6N hydrochloric acid for 24 hr under nitrogen. Samples for tryptophan were hydrolyzed with methanesulfonic acid. The amino acid residues were calculated as grams per 100 g of crude protein, and amino acids are reported as grams of the individual amino acid residue per 100 g of the total amino acid residue.

### INITIAL EQUATIONS UNSATISFACTORY

Amino acid analyses and PER values corrected to 2.50 casein were correlated to determine the relationships between PER and specific amino acids. Simple correlations between individual amino acids and PER ranged from 0.991 (leucine) to -0.690 (arginine); it is interesting that most of the simple correlations were greater than 0.90.

Due to the limitation of matrix size, only six amino acids could be used in the preliminary regression equation; thus the amino acids correlating most highly with PER were used to derive the first regression equation. A second regression equation was derived using the six nonessential amino acids correlating most highly with PER. The multiple correlation coefficients for these two regressions were 0.986 and 0.990, respectively.

The t values for the regression coefficients in these equations were quite low, which indicated that only leucine and proline were significant variables. There was considerable multiple colinearity in the equation due to the high correlations among the independent variables (Table 1); thus the first equations were unsatisfactory (Draper and Smith, 1966).

#### SATISFACTORY EQUATIONS OBTAINED

A multiple regression was then performed, setting leucine and proline as the independent variables with PER as the dependent variable. Leucine (LEU) and proline (PRO) were selected for this regression because they had a high positive correlation and a high negative correlation, respectively, with PER and also because they had a low simple correlation (-0.571) with each other. The multiple regression gave the following equation:

$$PER = -0.684 + 0.456(LEU) - 0.047(PRO) (1)$$

The multiple correlation coefficient for regression was 0.992; the coefficient of variation was 98.41%; and the F value of 145.2 for this regression equation indicated significance at the 99.5+% level. The t values for the regression coefficients showed that proline was not a significant variable; thus, leucine could be used alone in a very simple regression equation.

The lecine-proline equation was used to estimate the PER values on meat, yeast, soybean, fish, and poultry products shown in Table 2, using amino acid data supplied by several food companies. The equation gave good results for meat and poultry products but showed some large differences between predicted and observed PERs for other protein sources.

Two more approaches were used to develop estimating equations from the initial beef data—a stepwise regression with backward elimination, and a straightforward stepwise regression. The results are shown in Table 3. These two stepwise regressions progressed in the same manner for the first three steps, each selecting methionine, leucine, and tyrosine as the independent variables. The t value used to test significance was determined to be 2.201 for this number of variables (12) and observations (17). The backward elimination process recalculates the t values after each step. After the third step, the t value of methionine was recalculated and found to be less than 2.201; thus, it was eliminated from the equation. There were no other

Table 1—(Continued)

	VAL	ALA	ARG	ASP	GLU	GLY	HYLYS	HYPRO	PRO	SER
HIS	0.650	-0.436	-0.218	0.582	0.727	-0.727	-0.481	-0.873	-0.800	0.528
ILE	0.265	-0.714	-0.214	0.714	0.714	-0.857	-0.618	-0.857	-0.786	0.593
LEU	0.340	-0.357	-0.571	0.500	0.500	-0.643	-0.837	-0.643	-0.571	0.222
LYS	0.269	-0.800	-0.291	0.946	0.946	-0.946	-0.778	-0.800	-0.873	0.679
MET	0.115	-0.873	-0.218	0.873	0.873	-0.873	-0.704	-0.727	-0.800	0.755
CYSH	0.999	-0.076	-0.756	0.227	0.378	-0.378	-0.577	-0.529	-0.454	0.078
PHE	0.340	-0.643	-0.286	0.786	0.786	-0.929	-0.691	-0.929	-0.857	0.519
TYR	0.269	-0.727	-0.218	0.873	0.873	1.000	-0.778	-0.873	-0.800	0.604
THR	0.192	-0.800	-0.291	0.946	0.946	-0.800	-0.630	-0.800	-0.873	-0.830
TRY	0.200	-0.886	-0.322	1.000	1.000	-0.886	-0.698	-0.886°	-0.967	0.920
VAL		0.076	-0.907	0.076	0.227	-0.277	-0.423	-0.378	-0.302	-0.078
ALA	0.076	. * <u></u> }-	0.071	-0.857	-0.714	0.714	0.546	0.571	0.643	-0.815
ARG	-0.907	0.071	-	-0.214	-0.357	0.214	0.473	0.357	0.429	-0.074
ASP	0.076	-0.857	-0.214		0.857	-0.857	-0.691	-0.714	-0.786	0.741
GLU	0.227	-0.714	-0.357	0.857		-0.857	-0.691	-0.857	-0.929	0.741
GLY	-0.277	0.714	0.214	-0.857	-0.857		0.764	0.857	0.786	-0.593
HYLYS	-0.423	0.546	0.473	-0.691	-0.691	0.764	-	0.655	0.582	-0.377
HYPRO	-0.378	0.571	0.357	-0.714	-0.857	0.857	0.655		0.929	-0.593
PRO	-0.302	0.643	0.429	-0.786	-0.929	0.786	0.582	0.929		-0.667
SER	-0.078	-0.815	-0.074	0.741	0.741	-0.593	-0.377	-0.593	-0.667	J.007

Table 4—(Continued)

			4—(Continu							
	Bornard Carlotter			Estimated PER			Difference in PER (estimated — observed)			
Sample No.	Characterizing ingredients <sup>b</sup>	Observed PER	Eq. 1	2	3	Eq. 1	2	3		
arine and	vegetable combinations						and a feet of the	-0.3		
1367	(F20, V35)	2.0	1.8	1.9	1.7	<b>⊸</b> 0.2	-0.1			
	(F20,V20)	3.1	2.0	2.2	1.9	-1.1	-0.9	-1.2		
6241		3.6	2.0	2.3	2.4	-1.6	-1.3	-1.2		
6243	(F25,V25)		1.9	2.0	2.3	-0.6	-0.5	-0.2		
6245	(F30,V35)	2.5	1.7	2.0	2.0	777				
	getables, fish, and rice combin	nations 1.8	2.3	2.4	1.9	+0.5	+0.6	+0.1		
6294	(P10,V30,R15,F10)	1.0	2.0	7						
egetables	(no meat or poultry)	0.5	0.9	1.3	0.9	+0.4	+0.8	+0.4		
1601	(V50)	0.5			1.0	+0.7	+0.8	+0.2		
0571	(V65)	0.8	1.5	1.6		-0.3	0	0		
1021	(V58,N5)	1.2	0.9	1.2	1.2		A 5 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4 4	+0.3		
1141	(V50,N3)	0.9	1.3	1.6	1.2	+0.4	+0.7			
1151	(V35,N5)	1,1	0.8	0.9	. 1.0	-0.3	-0.2	-0.1		
		noultry)								
	dairy products (no meat or	2.4	1.6	1.9	2.2	-0.8	-0.5	-0.2		
2512	(N20,V15,D5)	2.4	0.3	2.0	2.4	-2.7	-0.4	0		
2701	(N15,D10)		2.4	2.7	2.3	-0.2	+0.1	-0.3		
6260	(N25,D15)	2.6			2.4	-0.2	-0.2	-0.1		
6265	(V20,N15,D10)	2.5	2.3	2.4	2.4	0.2	<b>U.</b> _			
/arious fo	od products with beans						+1.6	+1.4		
1377	(M10,B35,V30)	0.7	2.2	2.3	2.1	+1.5		+0.6		
		0.7	1.9	2.0	1.3	+1.2	+1.3			
1467	(M4,B50)	1.2	2.1	2.3	2.1	+0.9	+1.1	+0.9		
1857	(M11,B55,V10)	1.0	2.7	2.9	2.6	+1.7	+1.8	+1.6		
2387	(M20,B35,V20)		3.3	3.4	4.2	+2.8	+2.9	+3.7		
2957	(B80,M1)	0.5				+0.7	+0.9	0		
1197	(B40,V5)	0.9	1.6	1.8	0.9		+1.7	-0.1		
1291	(M5,B60)	0.7	2.3	2.4	0.6	+1.6				
	(M15,V15,B20)	1.7	2.4	2.6	1.9	+0.7	+0.9	+0.2		
6272 6292	(M12,B30,V25)	1.5	2.0	2.1	1.8	+0.5	+0.6	+0.3		
		* 3								
	ous products	2.8	2.9	2.9	2.9	+0.1	+0.1	+0.1		
1	Lean beef	2.0								
2	Partially defatted	0.4	2.3	2.5	2.3	-0.1	+0.1	-0.1		
**	chopped beef <sup>c</sup>	2.4	2.3	2.5	2.0	- Fig. 314				
3	Partially defatted	1 2	1.7	1.6	1.7	+0.1	+0.1	+0.1		
-	chopped beef	1.6	1.7	1.0		8 7				
4	Partially defatted cured chopped beef	2.6	2.7	2.1	2.6	+0.1	-0.5	0		
z <b>5</b>	Partially defatted beef	** 2					10.1	+0.2		
, <b>J</b>	fatty tissue°	1.1	1.3	1.2	. 1.3	+0.2	+0.1	10.2		
6	Partially defatted beef					-0.2	-0.3	+0.2		
	fatty tissue <sup>e</sup>	1.7	1.5	1.4	1.5	0.2	0.0			
	Partially defatted beef				1.5	-0.2	-0.3	-0.2		
	fatty tissue <sup>c</sup>	1.7	1.5	1.4	1.5		+0.3	-0.2		
8	Collagen <sup>e</sup>	0.0	0.1	0.3	0.2	+0.1	The second second second	+0.2		
		2.1	3.0	2.2	2.3	+0.9	-0.1			
9 /	Yeast protein	2.2	3.2	2.4	2.1	+1.0	+0.2	-0.		
10	Yeast protein	2.3	3.3	2.5	2.6	-1.0	+0.2	+0.3		
11	Yeast protein		2.6	2.0	1.8	+0.7	+0.1	-0.		
.12	Yeast cells	1.9			1.8	+0.6	-0.2	0		
13	Yeast cells	1.8	2.4			+0.8	+0.1	0		
14	Yeast cells	1.7	2.5	1.8	1.7		+0.1	-0.		
	Yeast cells	1.8	2.6	1.9	1.7	+0.8		0.		
15		2.1	2.0	2.0	2.1	+0.1	-0.1	v		
,16	Danish pastry						- 1	_		
17	Beef and partially defa beef fatty tissue	2.5	2.5	2.4	2.5	0	-0.1	0		
18			27. *		0.5	0	-0.1	0		
10	beef fatty tissue	2.5	2.5	2.4	2.5	U	0.1			

<sup>\*</sup> Data supplied by Campbell Soup Co., Anheuser Busch, Inc., J.M. Smucker Co., and Eastern Regional Research Center, USDA

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\* M = meat, P = poultry, F = marine products, D = dairy products, E = eggs, V = vegetables, N = noodles, R = rice, and B = beans. Thus, the notation (M20,V15,N20) indicates 20% meat, 15% vegetables, and 20% noodles; the unaccounted-for ingredient is either sauce, gravy, or brine used in the derivation of equations

# Equations Predict PER from Amino Acid Analysis . . .

Table 4—EFFECTIVENESS OF EQUATIONS in predicting PER of various food products\*

Sample	Characterizing ingredients <sup>b</sup>	Observed			Estimated PER			Diff (estim	ference in ated – ot	PER bserved)
No.		PER		Eq. 1	2	3		Eq. 1	2	3
Meat and	vegetable combinations									
1827	(M10V40)	1.7		1.4	1.6	1.6	. 3. 3	-0.3		Brook paragraps
2385	(M20V50)	2.3		2.6	2.8	2.3			-0.1	-0.1
0570	(M12V50)	2.0						+0.3	+0.5	0
1231	(M10V40)			3.5	3.6	4.0		+1.5	+1.6	+2.0
1287	(M10V35)	1.6		1.8	1.7	1.6		+0.2	+0.1	0
6210		1.6		1.6	1.9	1.7		0	+0.3	+0.1
	(M25V40)	3.0		2.8	2.8	3.2		-0.2	-0.2	100
6216	(M25V40)	3.1		2.8	2.8	2.9				
6230	(M30V35)	3.0		2.9				-0.3	-0.3	-0.2
6250	(M25V40)	2.8			2.6	3.0		-0.1	-0.4	0
6270	(M25V55)			2.8	2.8	2.8		0	0	0
6285	(M25V35)	2.9		1.7	2.8	2.5		-1.2	-0.1	-0.4
		2.5		2.3	2.3	2.6		-0.2	-0.2	
6316	(M25V30)	2.2		2.0	1.9	2.0		-0.2		+0.1
6321	(M20V25)	2.5		2.4	2.5				-0.3	-0.2
6341	(M20V20)	3.0				2.5		-0.1	0	0
6679	(M50V50)			2.1	2.8	2.7		-0.9	-0.2	-0.3
		2.5		2.4	2.4	2.7		-0.1	-0.1	+0.2
oultry and	vegetable combinations		*		*					
2386	(P15V50)	2.0		1.0	2.1					
0553	(P11V55)			1.9	2.1	2.0		-0.1	+0.1	0
1081	(P5V25)	1.8		1.9	2.0	1.8		+0.1	+0.2	Ō
6220		1.5		0.6	1.3	1.2		-0.9	-0.2	77.14
	(P45V40)	2.7		1.9	2.5	2.8		-0.8		-0.3
6290	(P20V40)	2.9		2.0	2.1	2.8			-0.2	-0.1
6311	(P30V35)	2.5		2.1				-0.9	-0.8	-0.1
6344	(P15V30)	2.7			2.3	2.3		-0.4	-0.2	-0.2
6677	(P50V45)			1.9	2.3	2.8		-0.9	-0.4	+0.1
6680	(P25V40)	2.7		2.2	2.3	2.6		-0.5	-0.4	-0.1
		2.6		2.0	2.1	2.3		-0.6	-0.5	-0.3
eat, noodl	e, and vegetable combinations							0.0	0.5	0.3
1651	(M15,V10,N12)	2 2			- B.					
2157	(M10,N10)	2.3		1.4	1.6	1.7		-0.9	-0.7	-0.6
		2.0		1.5	1.6	1.5		-0.5		
1137	(M15,V30,N5)	1.2		1.2	1.6	1.2			-0.4	-0.5
1221	(M10,N10,V5)	2.5						0	+0.4	0
6012	(M20,V15,N20)			1.6	1.8	2.2		-0.9	-0.7	-0.3
6113	(M20,V20,N10)	2.8		3.8	3.9	3.2		+1.0	+1.1	+0.4
6235	(M25, 120, 1470)	2.8		2.4	2.5	2.4	15 By	-0.4	-0.3	-0.4
6261	(M25,N20,V30)	2.6	200	2.6	2.7	2.7		0.4		
	(M10,V20,N20)	1.8		1.8	1.9	1.6			+0.1	+0.1
6291	(M10,N20,V20)	2.4		2.3			1.45	0	+0.1	-0.2
6678	(M25,N25,V40)	3.1			1.6	2.4		-0.1	-0.8	0
. 12				2.9	2.1	2.9		-0.2	-1.0	-0.2
ultry, vege	etable, and noodle combinations	:						JA Ball		
1541	(P7,N6,V5)	1.8		1 4	1.0					
1621	(P6,N10,V5)	2.2		1.6	1.8	1.6		-0.2	0	-0.2
0572	(P13,N6,V10)			1.1	1.3	1.9		-1.1	-0.9	-0.3
1071	(P7 NE 1/00)	1.9		1.2	1.4	1.8		-0.7	-0.5	
	(P7,N5,V80)	1.8		1.1	1.3	1.6				-0.1
1241	(P5,N15,V10)	2.5		1.3	1.5		3 2 ( )	-0.7	-0.5	-0.2
1251	(P5,N15,V2)	2.2				1.6		-1.2	-1.0	-0.9
1311	(P6,V30,N5)			1.5	1.7	2.2		-0.7	-0.5	0
5032	(P15,V15,N20)	1.9		1.2	1.3	2.0		-0.7	-0.6	+0.1
5092	(P15,V15,N20)	2.6		2.0	2.2	2.8		-0.6	-0.4	
5133	(D20 V20 V20	2.8		2.4	2.5	2.3				+0.2
	(P20,V20,N10)	2.6		3.1	3.0				-0.3	-0.5
5193	(P18,V15,N10)	2.8		2.4		2.6		+0.5	+0.4	0
5271	(P10,V20,N20)	2.9			2.5	2.4			-0.3	-0.4
		<i>2</i>		2.1	2.2	2.9			-0.7	0
at and dai	ry products combinations								wy in the	··
2540	(M12,N12,D2)	2.5		2 1 1	^ ~			354		
274	(M20,V35,D5)	2.9		2.1	2.0	2.3	•	-0.3	-0.5	-0.2
4/ 1-		4.7		2.0	2.2	2.7			-0.7	-0.2
t/poultry	and egg combinations									J.4.
841	(P6,E6,V5)	2.0		1.0						
686	(M30,E20)			1.9	2.1	2.1	er ja er	-0.1	+0.1	+0.1
690	(M20,E10)	2.8		2.6	2.7	2.7			+0.1	
-,0	(M15,E50)	2.9		2.8	2.4	2.9			+0.1 −0.5	-0.1 0
695										

## Equations Predict PER from Amino Acid Analysis . . .

Table 2—EFFECTIVENESS OF EQUATION 1 in estimating PER of various foods

Food product	Estimated PER using Equation 1	Observed PER	Difference
Lean beef*	2.90	2.85	0.05
Partially defatted			
chopped beef*	2.33	2.38	0.05
Partially defatted			
chopped beef*	1.68	1.61	0.07
Partially defatted	en de la companya de La companya de la co		
cooked cured beef*	2.65	2.58	0.07
Partially defatted			
beef fatty tissue	1.26	1.13	0.13
Partially defatted			
beef fatty tissue*	1.52	1.70	0.18
Meat and vegetables	1.7	1.4	0.3
Meat and vegetables	2.5	2.3	0.2
Meat and vegetables	1.6	1.6	0
Poultry and vegetables	1.9	2.0	0.1
Poultry and vegetables	1.9	1.8	0.1
Poultry and vegetables	2.4	2.2	0.2
Meat, noodles and			
vegetables	2.9	3.1	0.2
Vegetables	1.5	0.8	0.7
Vegetables and noodles	1.3	0.9	0.4
Meat and beans	2.2	0.7	1.5
Meat and beans	2.8	0.8	2.0
Poultry and beans	1.0	2.6	1.6
Noodles and cheese	0.3	2.4	2.7
Noodles and cheese	2.4	2.6	0.2
Noodles, vegetables,			
and cheese	1.6	2.4	0.8

<sup>\*</sup> Foods used to develop Equation 1

Table 3—CHARACTERISTICS OF THE STEPWISE REGRES-SION, with and without backward elimination

Variable	Regression coefficient	t value	F value
With backward elimination			
Leucine	0.454	6.64	106.67
Tyrosine	-0.105	4.53	17.25
Intercept	-0.468		
Standard error of regression	0.23		
Multiple correlation	0.949		
Coefficient of determination	0.8931		
t value required			
for significance	2.201		
F value required			
for significance	2.50		
Without backward elimination			
Methionine	0.435	3.34	42.73
Leucine	0.780	4.90	6.87
Tyrosine	-0.944	4.24	13.26
Histidine	0.211	2.37	5.65
Intercept	-1.816		
Standard error of regression	0.16		
Multiple correlation	0.978		
Coefficient of determination	0.9564		
t value required			
for significance	2.201		•
F value required			
for significance	2.50		

variables with t values greater than 2.201, so the regression was terminated with two variables—leucine and tyrosine—to give the following equation:

$$PER = -0.468 + 0.454(LEU) - 0.105(TYR)$$
 (2)

The straightforward stepwise regression calculates t values only at the start of the regression and then proceeds to add all variables with t values greater than the minimum value needed for significance (i.e., 2.201). In the straightforward regression, there were four variables with t values greater than 2.201: methionine, leucine, tyrosine, and histidine. The resulting equation is as follows:

$$PER = -1.816 + 0.435(MET) + 0.780(LEU) + 0.211(HIS) - 0.944(TYR)$$
(3)

The t and F values for Equations 2 and 3 indicate that they are both valid, with significant variables. The four-variable equation (Eq. 3) has a higher coefficient of determination and explains 95.6% of all deviation from regression, compared with 89.3% for Equation 2.

### TESTING THE EQUATIONS

Keep in mind that regression analysis is only a mathematical analysis that determines the mathematical relationship between a dependent variable (PER in this case) and one or more independent variables. There is no causal relationship determined or implied. The four amino acids selected in Equation 3 (methionine, leucine, histidine, and tyrosine) are not to be regarded as having more biological significance than other amino acids present, but only as having a more significant mathematical relationship to PER. In other words, the changes in amounts of these amino acids have value in predicting the changes in PER, but may not have a direct relation to PER.

A regression equation can only be assumed to be linear over the range covered by the values of the variables used in the regressions. The values in this study were:

Histidine	0.8- 3.8%
Leucine	3.8- 9.3%
Methionine	0.7- 2.7%
Tyrosine	0.9- 4.8%
Proline	3.6-13.8%

The prediction equations were tested on amino acid-PER data supplied by several food corporations. In previous studies, the standard error among duplicate PER determinations was shown to be 0.2 PER, which indicates that no difference exists between PER values that differ by 0.2 or less. Therefore, the criteria for acceptance of a prediction equation was set at  $\pm 0.2$  PER.

Table 4 (p. 38) presents the results of testing the effectiveness of the three equations to predict the PER of various food products. Equation 3 is a reliable estimator of PER when it is used with products that contain proteins primarily of meat, poultry, grain, or yeast origin. This equation successfully predicted the PER  $(\pm 0.2)$  of 66 out of 93 food products—an effectiveness of 71%. Equations 1 and 2 were ineffective in predicting PER of foods containing little or no meat or poultry.

In some instances (e.g., products containing fish or beans), all three equations failed to predict PER with accuracy; and the data suggest that extreme caution should be exercised in estimating PER for such prod-

### Equations Predict PER from Amino Acid Analysis . . .

ucts. For two foods containing marine products, the equations somewhat underestimated the PER, while for those products containing beans, the equations greatly overestimated the PER. These errors could be attributed to differences in the digestibility of these type of proteins and the fact that the amino acid profiles of these unknown mixtures may vary considerably from the proteins of meat that were used to develop the equation. For instance, beans are high in leucine, an amino acid of greatest importance in these equations. Additional work must be done on these products to establish regression relationships between PER and the amino acids of bean, marine, and noodle products.

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